Journal of Oral Science, Vol. 52, No. 4, 633-640, 2010

Original

Aberrant β-catenin expression in the histologic differentiation of oral squamous cell carcinoma and verrucous carcinoma: an immunohistochemical study

Lankesh B. Laxmidevi[1)], Punnya V. Angadi[2)], Rekha K. Pillai[3)] and Chethana Chandreshekar[4)]

[1)]Department of Oral and Maxillofacial Pathology, Sri Siddhartha Dental College and Hospital, Tumkur, India
[2)]Department of Oral Pathology, KLE VK Institute of Dental Sciences and Hospital, Belgaum, Karnataka, India
[3)]Shri Dharmasthala Manjunathehwara College of Dental Sciences and Hospital, Dharwad Karnataka, India
[4)]College of Dental Surgery, Manipal, Karnataka, India

(Received 20 July and accepted 1 Novembre 2010)

Abstract: β-Catenin acts as a structural protein at cell-cell adherens junctions and as a transcription activator mediating Wnt signal transduction. Altered β-catenin expression has been associated with loss of cell differentiation and acquisition of an invasive phenotype. In the present study, β-catenin expression was compared immunohistochemically between oral squamous cell carcinoma (30 cases) and verrucous carcinoma (30 cases), and correlated with different histological grades of oral squamous cell carcinoma. Positivity for β-catenin was seen in 17 cases (56.6%) of oral squamous cell carcinoma and 25 cases (83.3%) of verrucous carcinoma, and was significantly correlated with the grade of oral squamous cell carcinoma, whereas no significant correlation of β-catenin expression was observed between oral squamous cell carcinoma and verrucous carcinoma. In oral squamous cell carcinoma, the number of β-catenin-positive cases and the intensity of expression decreased as cancers became more poorly differentiated. Decreased membranous localization and intense cytoplasmic staining were observed in poorly differentiated squamous cell carcinoma. In verrucous carcinoma, β-catenin was demonstrable mainly in the membrane. Down-regulation of β-catenin was significantly correlated with lack of differentiation in oral squamous cell carcinoma. Reduced membranous expression and predominant cytoplasmic localization were prominent among higher-grade tumors, suggesting stabilization of β-catenin and its role as a signaling molecule. Predominant membranous expression in verrucous carcinoma was similar to that observed in well differentiated squamous cell carcinoma, thus corroborating its role in cell adhesion in these subgroups. (J Oral Sci 52, 633-640, 2010)

Keywords: β-catenin; oral squamous cell carcinoma; verrucous carcinoma; histologic differentiation.

Correspondence to Dr. Punnya V. Angadi, Department of Oral Pathology, KLE VK Institute of Dental Sciences and Hospital, Nehru Nagar, Belgaum 590010, Karnataka, India
Tel: +98-80845530
Fax: +98-831-2470640
E-mail: punnya_angadi@rediffmail.com

Introduction

Cell adhesion molecules play important roles in cell development, differentiation, induction of cellular morphogenesis and maintenance of structural integrity (1). They also participate in a large variety of signal transduction events that are important for regulating cell adhesion, motility, growth, apoptosis and specific gene regulation (2). Consequently, molecular mechanisms of cell adhesion have been considered to be key components in the metastatic cascade, involving many interactions between tumor and host cells in angiogenesis, proteolysis, motility and invasion (3,4). β-Catenin, a 92-kDa protein, plays

different roles in the cell as both a structural protein at cell-cell adherens junctions and a transcriptional activator mediating Wnt signal transduction (5). β-Catenin forms complexes with E-cadherin and α-catenin to establish links with the actin cytoskeleton. Any disruption of β-catenin binding, or deletion of the gene, leads to loss of cell-cell adhesion and disorganization of cells (6,7).

Nonjunctional/free β-catenin is sequestered in a complex with the adenomatous polyposis coli (APC) molecule, and with GSK3β (serine-threonine glycogen synthetase kinase), along with an adapter protein, axin, enabling phosphorylation and degradation of β-catenin by the ubiquitin proteasome system (8,9). This is regulated by the Wnt signaling pathway, which plays a central role in cell adhesion, proliferation, differentiation, and epithelial mesenchymal transition (10,11). Activation of Wnt signaling increases the stability of β-catenin by inactivating GSK3β, and as a consequence, the cytoplasmic level of β-catenin increases (12). This stabilization of cytoplasmic β-catenin results in an increased nuclear level of β-catenin, leading to formation of a complex with the T cell factor/lymphoid enhancer factor (TCF/LEF) family of transcription factors, which recruit components of the transcriptional machinery and cause activation of target genes (12,13) including, for example, cyclin D1, a critical cell cycle regulator (14-16). These mutations stimulate cell proliferation and the development of various neoplasms (12). Furthermore, loss or down-regulation of β-catenin protein at the membrane has been associated with reduced intercellular adhesion (16). Alteration of β-catenin expression has thus been associated with loss of differentiation and acquisition of an invasive phenotype in a range of tumors (17-23). Several studies have evaluated the expression of β-catenin in oral squamous cell carcinomas (OSCC) (24-30), but no such studies have focused on verrucous carcinoma (VC). Therefore, the present study was undertaken to evaluate the expression of β-catenin in these two neoplasms to assess its possible role in tumor progression, differentiation and behavior.

Materials and Methods

This laboratory-based study involved the use of buffered formalin-fixed, paraffin-embedded tissues of histopathologically diagnosed cases of OSCC and VC retrieved from the files of the Department of Oral Pathology and Microbiology, Shri Dharmasthala Manjunatheswara College of Dental Sciences, Dharwad, Karnataka. The study protocol was approved by the institutional ethics committee.

A total of 60 cases were evaluated immunohistochemically for β-catenin expression. These included 30 cases of OSCC i.e. 10 cases each of well differentiated (WDSCC), moderately differentiated (MDSCC) and poorly differentiated (PDSCC) squamous cell carcinomas and 30 cases of VC. The diagnosis of VC was established on the basis of an exophytic tumor with endophytic proliferation of minimally dysplastic epithelium in the form of broad bulbous rete ridges which extended below the level of the surrounding mucosa. The surface usually exhibited hyper-keratinization and clefts plugged with keratin (31). The diagnosis was confirmed by two oral pathologists using sections stained with hematoxylin and eosin.

Immunohistochemistry

Two or three serial sections 4 μm thick were prepared and placed on silanized slides. The sections were deparaffinized and rehydrated through xylene and descending grades of alcohol. Antigen retrieval was carried out in a pressure cooker in 10 mM citrate buffer (pH 6.0) for 2 to 5 min.

The sections were then incubated after covering them with 3% hydrogen peroxide for 15 min to block any endogenous peroxidase activity, and then incubated with primary anti-β-catenin monoclonal antibody (DAKO Cytomation, USA, clone β-catenin-1) for 4 hours at room temperature using an optimal dilution of 1:50.

After further incubation with the secondary antibody (45 min) and streptavidin peroxidase (30 min), visualization was performed using freshly prepared diaminobenzidine (DAB) chromogen for 10 min. The slides were finally counterstained with Harris hematoxylin.

For each batch of slides, a negative control in which the primary antibody was replaced by Tris-buffered saline and a positive control of normal epithelium (Fig.1) were used. A brown-colored product on the membrane, cytoplasm and nucleus was considered to be indicative of positivity. The slides were assessed for staining intensity, which was graded as mild (1), moderate (2), or intense (3), and the localization of the antigen was also analyzed. Two other observers (the second and third authors) evaluated the slides to eliminate any interobserver bias.

A rank sum two-sample Mann-Whitney *U*-test was used for comparison and correlation between OSCC and VC and between different grades of OSCC.

Results

Normal epithelium that had been used as a control exhibited strong homogeneous β-catenin expression at the membranes (Fig. 1). Positivity for β-catenin was seen in 17 cases (56.6%) of OSCC and 25 cases (83.3%) of VC. Among the OSCCs, WDSCC was positive in 10/10 cases

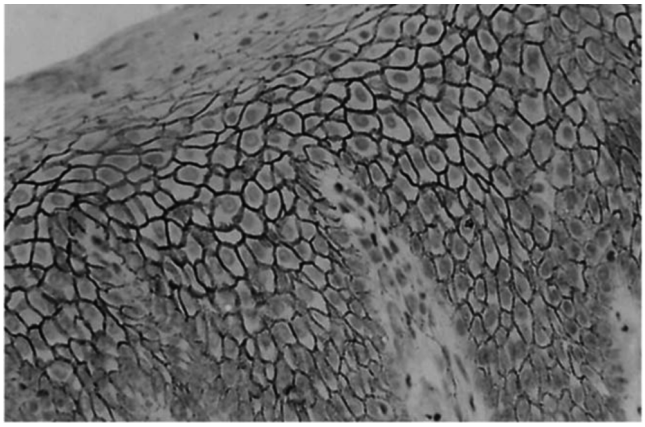

Fig. 1 Normal epithelium demonstrating intense membranous expression (× 250).

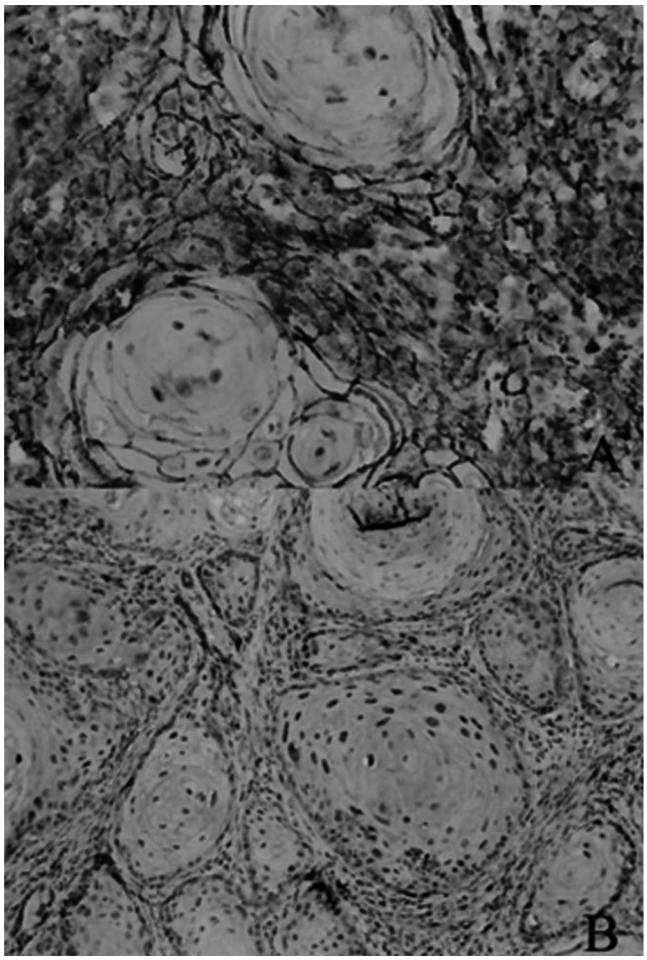

Fig. 2 A: Intense membranous β-catenin expression in well differentiated squamous cell carcinoma (× 250). B: Nuclear β-catenin expression in well differentiated squamous cell carcinoma.

(Fig. 2A and B), MDSCC in 4/10 cases (Fig. 3A) and

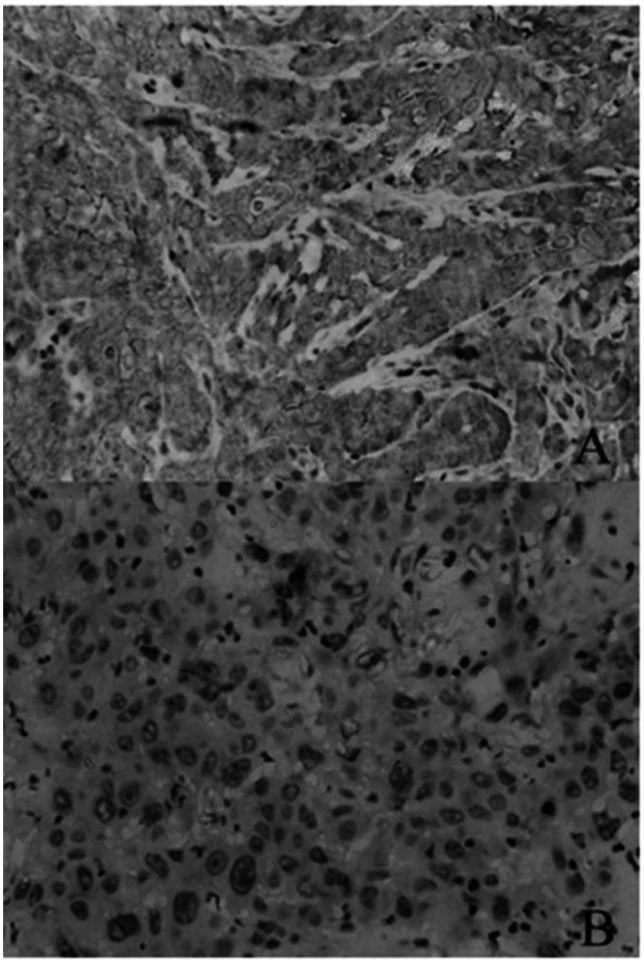

Fig. 3 A: Cytoplasmic β-catenin expression in moderately differentiated squamous cell carcinoma. B: Lack of staining observed in poorly differentiated squamous cell carcinoma.

PDSCC in 3/10 cases (Fig. 3B). The grades of staining intensity are shown in Table 1. Table 2 shows the localization of β-catenin expression in the different groups of carcinomas. The number of positive cases and the intensity of staining decreased with increasing grade in OSCC, whereas in VC 11 cases were mildly positive and 7 cases each were moderately and intensely positive for β-catenin.

Regarding the localization of β-catenin, a decrease of membranous staining and intense cytoplasmic staining were observed in PDSCC relative to WDSCC and MDSCC. In VC, β-catenin was demonstrable mainly in the membrane (Figs. 4A and B).

There was a significant ($P < 5$) correlation of β-catenin expression between MDSCC and PDSCC, and between WDSCC and PDSCC. However, there was no such significant correlation between OSCC and VC (Table 3).

Table 1 Positivity for β-catenin and its degree of intensity in different groups of oral squamous cell carcinomas and verrucous carcinoma

Group of cases	Total cases	Positive cases	Percentage	Intensity of staining		
				Mild	Moderate	Intense
OSCC	30	17	56.7%	4	4	9
WDSCC	10	10	100%	1	3	6
MDSCC	10	4	40%	0	1	3
PDSCC	10	3	30%	3	0	0
VC	30	25	83.3%	11	7	7

OSCC: Oral squamous cell carcinoma; WDSCC: well differentiated squamous cell carcinoma; MDSCC: moderately differentiated squamous cell carcinoma; PDSCC: poorly differentiated squamous cell carcinoma; VC: Verrucous carcinoma.

Table 2 Localization of β-catenin in the various groups of tumors studied

Group of cases	Positive cases	Membrane	Cytoplasm	Nuclear
OSCC	17	12	4	1
WDSCC	10	9	0	1
MDSCC	4	3	1	0
PDSCC	3	0	3	0
VC	25	25	0	0

OSCC: Oral squamous cell carcinoma; WDSCC: well differentiated squamous cell carcinoma; MDSCC: moderately differentiated squamous cell carcinoma; PDSCC: poorly differentiated squamous cell carcinoma; VC: Verrucous carcinoma

Table 3 Statistical analyses of differences in β-catenin immunostaining in various groups of tumors

Group	Total cases	Positive staining				
		Mild	Moderate	Intense	Negative	*P* value
WDSCC	10	1	3	6	0	
MDSCC	10	0	1	3	6	0.0494 (S)
WDSCC	10	1	3	6	0	
PDSCC	10	3	0	0	7	0.0003 (S)
MDSCC	10	0	1	3	6	
PDSCC	10	3	0	0	7	0.4274 (NS)
OSCC	30	4	4	9	13	
VC	30	11	7	7	5	0.3871 (NS)

OSCC: Oral squamous cell carcinoma; WDSCC: well differentiated squamous cell carcinoma; MDSCC: moderately differentiated squamous cell carcinoma; PDSCC: poorly differentiated squamous cell carcinoma; VC: Verrucous carcinoma
NS: not significant; S: significant

Discussion

Adhesion between adjacent epithelial cells and between cells and extracellular matrix plays a significant role in tissue morphogenesis in embryos and in the maintenance of complex differentiated tissues in adults (8). Reduced cell-cell adhesion is associated with loss of contact inhibition for proliferation, thereby allowing escape from growth control signals, and triggering carcinogenesis (32,33).

β-Catenin, an important component of the E-cadherin-catenin adhesion complex, plays a dual role (34,35) as a structural protein at cell-cell adherens junctions and as a

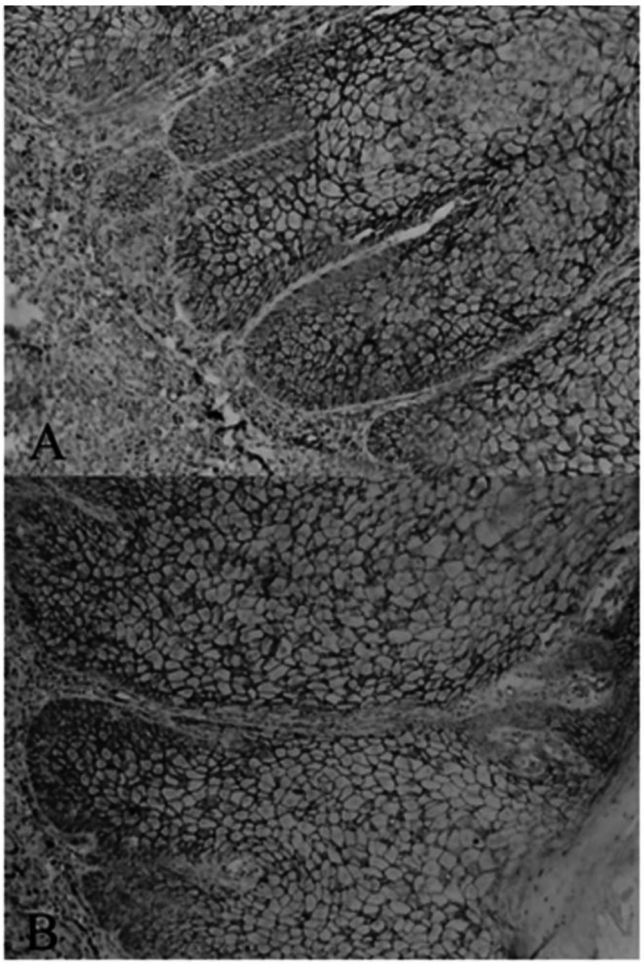

Fig. 4 Intense membranous expression of β-catenin in verrucous carcinoma (A: × 100) (B: × 250).

transcriptional activator mediating Wnt signal transduction (10,36,37). Altered/reduced β-catenin expression has been associated with loss of differentiation and acquisition of an invasive phenotype. Such altered β-catenin expression can be attributed to mutated APC gene products, and also mutations in the axin 1 and β-catenin genes. These mutations can lead to decreased degradation of β-catenin, abnormal phosphorylation, and activation of receptor kinases, with the consequence that β-catenin becomes unable to bind to E-cadherin (5,37).

In various studies, altered/reduced expression of β-catenin has been correlated with histological grade, an aggressive phenotype, prognosis, and an increased risk of invasion and metastasis (20,38,39). In a study of head and neck squamous cell carcinoma, β-catenin was suggested to function mainly as an adhesive, and not as a signaling, molecule (4). It has been suggested that β-catenin is a potent oncogene product (33), and its accumulation has been implicated in tumorigenesis in various tumors, although decreased expression has also been found in esophageal, colon, gastric, breast, prostate and oral cancers (21,27,40,41).

In the present study, levels of immunohistochemical expression of β-catenin were analyzed in OSCCs of different grades – WDSCC (10 cases), MDSCC (10 cases) and PDSCC (10 cases) – and also in 30 cases of VC, to elucidate its role in these tumors.

We found that 56.6% of OSCCs were positive for β-catenin, in accordance with studies done by William et al., (24) and Baggutti et al., (27), who reported corresponding figures of 66.6% and 68.2%, respectively. However, a high percentage of positivity (80%) was reported by Gao et al., (29).

In the present study, 43.3% of OSCC cases did not express β-catenin, which was similar to figures reported by William et al., (33.3%) (24) and Baguti et al., (31.8%) (27), but in contrast to that of Gasporoni et al. who did not report any negative expression. (28) These variations might have been attributable to the fixation process, the type of antibody (monoclonal/polyclonal), or the immunohistochemistry procedure employed. Our study employed a proper protocol with appropriate positive and negative controls with each batch of staining.

Among the different grades of OSCC, all (100%) of the cases of WDSCC, 4 (40%) of the MDSCCs and 3 (30%) of the PDSCCs showed positivity for β-catenin expression. This was in accordance with the study of Ueda et al., (30), who found that 80% of WDSCCs, 66.66% of MDSCCs and 40% of PDSCCs were positive.

A proportional decrease in the number of positive cases with increasing histopathologic grade was observed. The decrease of positivity seen in PDSCC could have been attributable to loss of the adhesive function of β-catenin and increased membranous degradation in more aggressive neoplasms.

Furthermore, in the positive cases, β-catenin expression was graded according to the intensity of staining as mild, moderate or intense, and compared among the groups. Intense staining was seen in 52.94% (9/17) of OSCCs, which appeared to be in accord with Kurtz et al., (25), who found intense staining in 55.55% of examined cases. Mild (23.52%) and moderate (23.52%) positivity for β-catenin in the present OSCCs was contradictory to the results obtained by Ueda et al., (30) and Kurtz et al., (25).

The staining intensity of β-catenin varied widely among the different grades of OSCC. In WDSCCs, the intensity of β-catenin expression varied from intense (60%) to moderate (30%) to mild (10%). These findings were comparable to those of Udea et al., (30), especially for intense (66.66%) and moderate (33.33%) staining. In

contrast, however, Gasporoni et al., (28) found predominantly mild staining (68.75%), and a lower extent of intense staining (31.25%).

In our present series, MDSCCs showed only intense (30%) and moderate (10%) staining intensity. Other studies have demonstrated varied expression of β-catenin in MDSCCs, being only intense (36.23%) in that of Ueda et al., (30), but predominantly mild (75%) in that of Gasporoni et al. (28), which was in contrast to our result. The PDSCCs in the present study demonstrated only mild staining intensity (30%) for β-catenin, which was not in accord with the study by Gasporoni et al., (28), who observed both intense (50%) and mild (50%) staining.

The intensity of β-catenin expression was correlated with the differentiation of OSCC, showing a decrease as the histological grade increased from WDSCC to PDSCC. This could have been attributable to increased protein degradation associated with PDSCC, resulting in reduced binding, and thus creating variations in the intensity (26,30).

With regard to the localization of β-catenin expression, predominant membranous expression was seen in most cases of WDSCC and MDSCC, being comparable with results obtained by Yu et al., (4), Gasporoni et al., (28) and Bagutti et al., (27). Predominant cytoplasmic staining of β-catenin in PDSCC in the present study was similar to that reported by William et al., (24) and Gao et al., (29). β-Catenin expression in the nucleus in WDSCC (1 case) and its cytoplasmic expression in a single case of MDSCC in the present series were also reported by Kurtz et al., (25). Predominant cytoplasmic staining may have been attributable to loss of β-catenin membrane localization due to impaired degradation of the protein, possibly due to mutations in the APC or β-catenin gene, resulting in accumulation of the protein in the cytoplasm. This would have led to β-catenin functioning inappropriately as a transcriptional activator, resulting in aggressive behavior of the tumor. Simultaneous expression in the membrane and cytoplasm suggests that the normal degradation of free β-catenin in the cytoplasm is inhibited. These mutations can also cause accelerated tumor cell proliferation through transcriptional activation of target genes such as cyclin D1, resulting in nuclear accumulation of β-catenin, as has been reported in a case of WDSCC (42). Thus, reduced membranous expression and increased cytoplasmic expression of β-catenin in PDSCC reflects the aggressive nature of the tumor. The significant correlation observed in the present study between the histological grade of the tumor and reduction of β-catenin expression supports its role as a prognostic marker

Verrucous carcinoma (VC) of the oral cavity should be considered as a clinicopathological entity distinct from the more common squamous cell carcinoma because of its unique biological behavior. This tumor shows much slower growth with a limited propensity to metastasize, thus having a better prognosis than OSCC. Significant clinical and biological differences between OSCC and VC have been found.

In the present study we found that β-catenin was expressed in 83.3% of VCs, with localization limited only to the membrane. Among these positive cases, 11 showed mild positivity (44%), 7 showed moderate positivity (28%), and 7 showed intense (28%) positivity. No expression was seen in 16.67% of the cases. This appears to be the first study of β-catenin expression in VC, and thus our results cannot be compared with previous findings.

Our study demonstrated some variation of β-catenin positivity in OSCC and VC, being 56.66% in the former and 83.33% in the latter. The staining intensity was predominantly mild in VC but not so mild in OSCC. With regard to localization, β-catenin expression was only membranous in VC (83.3%), whereas in OSCC membrane staining accounted for only around 40% of cases. This might reflect the difference in prognosis and behavior of these two carcinomas. However, these differences in the localization of immunostaining were not statistically significant.

Positivity for β-catenin was observed in 100% of WDSCCs as compared with 83.3% of VCs. The intensity of β-catenin expression in VC was predominantly mild (44%), unlike WDSCC, in which the staining was predominantly intense (60%). This difference in the intensity and positivity of staining may have been attributable to the relatively small number of WDSCC samples examined. Further study with a larger sample size would be of help for comparing the two neoplasms, which are thought to have similar behavior. However, the predominant membranous expression in VC was similar to that in WDSCC, suggesting that β-catenin still plays a cell-adhesive role in these subgroups.

Down-regulation of β-catenin expression was correlated with a poor histological grade of OSCC. The reduced expression associated with increased cytologic grade probably reflects the intense proliferative activity and invasiveness of these lesions. In addition, however, our results suggest that the level of β-catenin affects the differentiation of the cells in these oral neoplasms, as decreased expression was correlated with a lack of differentiation. Reduced membranous expression and predominant cytoplasmic localization were prominent among the higher-grade tumors, suggesting stabilization of β-catenin and its role as a signaling molecule. The

predominant membranous expression in VC was similar to that observed in WDSCC, corroborating the role of β-catenin in cell adhesion in these subgroups.

References

1. Blanco D, Vicent S, Elizegi E, Pino I, Fraga MF, Esteller M, Saffiotti U, Lecanda F, Montuenga LM (2004) Altered expression of adhesion molecules and epithelial-mesenchymal transition in silica-induced rat lung carcinogenesis. Lab Invest 84, 999-1012.
2. Gumbiner BM (1996) Cell adhesion: the molecular basis of tissue architecture and morphogenesis. Cell 84, 345-357.
3. Choi YL, Xuan YH, Shin YK, Chae SW, Kook MC, Sung RH, Youn SJ, Choi JW, Kim SH (2004) An immunohistochemical study of the expression of adhesion molecules in gallbladder lesions. J Histochem Cytochem 52, 591-601.
4. Yu Z, Weinberger PM, Provost E, Haffty BG, Sasaki C, Joe J, Camp RL, Rimm DL, Psyrri A (2005) β-Catenin functions mainly as an adhesion molecule in patients with squamous cell cancer of the head and neck. Clin Cancer Res 11, 2471-2477.
5. Zhurinsky J, Shtutman M, Ben-Ze'ev A (2000) Plakoglobin and β-catenin: protein interactions, regulation and biological roles. J Cell Sci 113, 3127-3139.
6. Pollack AL, Barth AI, Altschuler Y, Nelson WJ, Mostov KE (1997) Dynamics of β-catenin interactions with APC protein regulate epithelial tubulogenesis. J Cell Biol 137, 1651-1662.
7. Oyama T, Kanai Y, Ochiai A, Akimoto S, Oda T, Yanagihara K, Nagafuchi A, Tsukita S, Shibamoto S, Ito F, Takeichi M, Matsuda H, Hirohashi S (1994) A truncated β-catenin disrupts the interaction between E- cadherin and α-catenin: a cause of loss of intercellular adhesiveness in human cancer cell lines. Cancer Research 54, 6282-6287.
8. Wijnhoven BPL, Dinjens WNM, Pignatelli M (2000) E-cadherin-catenin cell-cell adhesion complex and human cancer. Br J Surg 87, 992-1005.
9. Salomon D, Sacco PA, Roy SG, Simcha I, Johnson KR, Wheelock MJ, Ben-Ze'ev A (1997) Regulation of β-catenin levels and localization by overexpression of plakoglobin and inhibition of the ubiquitin-proteosome system. J Cell Biol 139, 1325-1335.
10. Queimado L, Obeso D, Hatfield MD, Yang Y, Thompson DM, Reis AM (2008) Dysregulation of Wnt pathway components in salivary gland tumors. Arch Otolaryngol Head Neck Surg 134, 94-101.
11. Nusse R (2005) Wnt signaling in disease and in development. Cell Res 15, 28-32.
12. Daa T, Kashima K, Kaku N, Suzuki M, Yokoyama S (2004) Mutations in components of the Wnt signaling pathway in adenoid cystic carcinoma. Mod Pathol 17, 1475-1482.
13. Gottardi CJ, Gumbiner BM (2001) Adhesion signaling: how β-catenin interacts with its partners. Curr Biol 11, R792-794
14. Conacci-Sorell M, Zhurinsky J, Ben-Ze'ev A (2002) The cadherin-catenin adhesion system in signaling and cancer. J Clin Invest 109, 987-991.
15. Shtutman M, Zhurinsky J, Simcha I, Albanese C, D'Amico M, Pestell R, Ben-Ze'ev A (1999) The cyclin D1 gene is a target of the beta-catenin/LEF-1 pathway. Proc Natl Acad Sci USA 96, 5522-5527.
16. Shiozaki H, Oka H, Inoue M, Tamura S, Monden M (1996) E-cadherin mediated adhesion system in cancer cells. Cancer 77, 1605-1613
17. Xu HT, Wang L, Lin D, Liu Y, Liu N, Yuan XM, Wang EH (2006) Abnormal β-catenin and reduced axin expression are associated with poor differentiation and progression in non-small cell lung cancer. Am J Clin Pathol 125, 534-541.
18. Lowy AM, Fenoglio-Preiser C, Kim OJ, Kordich J, Gomez A, Knight J, James L, Groden J (2003) Dysregulation of β-catenin expression correlates with tumor differentiation in pancreatic duct adenocarcinoma. Ann Surg Oncol 10, 284-290.
19. Karayiannakis AJ, Nakopoulou L, Gakiopoulou H, Keramopoulos A, Davaris PS, Pignatelli M (2001) Expression patterns of β-catenin in-situ and invasive breast cancer. Eur J Surg Oncol 27, 31-36.
20. Wong NA, Pignatelli M (2002) β-Catenin – a linchpin in colorectal carcinogenesis? Am J Pathol 160, 389-401.
21. Takayama T, Shiozaki H, Shibamoto S, Oka H, Kimura Y, Tamura S, Inoue M, Monden T, Ito F, Monden M (1996) β-Catenin expression in human cancers. Am J Pathol 148, 39-46.
22. Krishnadath KK, Tilanus HW, van Blankenstein M, Hop WC, Kremers ED, Dinjens WN, Bosman FT (1997) Reduced expression of the cadherin-catenin complex in oesophageal adenocarcinoma correlates with poor prognosis. J Pathol 182, 331-338.
23. Faleiro-Rodrigues C, Masedo-Pinto I, Periera D, Ferriera VM, Lopes CS (2004) Association of E-cadherin and beta-catenin immuno-expression with clinicopathologic features in primary ovarian carcinomas. Human Pathol 35, 663-669.

24. William HK, Sanders DSA, Jankowski JAZ, Landini G, Brown AM (1998) Expression of cadherins and catenins in oral epithelial dysplasia and squamous cell carcinoma. J Oral Pathol Med 27, 308-317.
25. Kurtz KA, Hoffman HT, Zimmerman MB, Robinson RA (2006) Decreased E-cadherin but not β-catenin expression is associated with vascular invasion and decreased survival in head and neck squamous carcinomas. Otolaryngol Head Neck Surg 134, 142-146.
26. Kudo Y, Kitajima S, Ogawa I, Hiraoka M, Sargolzaei S, Keikhaee MR, Sato S, Miyauchi M, Takata T (2004) Invasion and metastasis of oral cancer cells require methylation of E-cadherin and/or degradation of membranous β-catenin. Clin Cancer Res 10, 5455-5463.
27. Bagutti C, Speight PM, Watt FM (1998) Comparison of integrin, cadherin, and catenin expression in squamous cell carcinomas of the oral cavity. J Pathol 186, 8-16.
28. Gasparoni A, Chaves A, Fonzi L, Johnson GK, Schneider GB, Squier CA (2002) Subcellular localization of β-catenin in malignant cell lines and squamous cell carcinomas of the oral cavity. J Oral Pathol Med 31, 385-394.
29. Gao S, Eiberg H, Krogdahl A, Liu CJ, Sørensen JA (2005) Cytoplasmic expression of E-cadherin and β-catenin correlated with LOH and hypermethylation of the APC gene in oral squamous cell carcinomas. J Oral Pathol Med 34, 116-119.
30. Ueda G, Sunakawa H, Nakamori K, Shinya T, Tsuhako W, Tamura Y, Kosugi T, Sato N, Ogi K, Hiratsuka H (2006) Aberrant expression of β- and γ-catenin is an independent prognostic marker in oral squamous cell carcinoma. Int J Oral Maxillofac Surg 35, 356-361.
31. Odell EW, Morgan PR (1998) Biopsy pathology of the oral tissues. Chapman and Hall Medical, Cambridge, 229-233.
32. Sadot E, Geiger B, Oren M, Ben-Ze'ev A (2001) Down-regulation of β-catenin by activated p53. Mol Cell Biol 21, 6768-6781.
33. Piepenhagen PA, Nelson WJ (1993) Defining E-cadherin-associated protein complexes in epithelial cells: plakoglobin, β- and γ-catenin are distinct components. J Cell Sci 104, 751-762.
34. Ilyas M, Tomlinson IPM, Rowan A, Pignatelli M, Bodmer WF (1997) β-Catenin mutations in cell lines established from human colorectal cancers. Proc Natl Acad Sci U.S.A. 94, 10330-10334.
35. Pötter E, Bergwitz C, Brabant G (1999) The cadherin-catenin system: implications for growth and differentiation of endocrine tissues. Endocr Rev 20, 207-239.
36. Shapiro L (2001) β-Catenin and its multiple partners: promiscuity explained. Nat Struct Biol 8, 484-487.
37. Hinck L, Näthke IS, Papkoff J, Nelson WJ (1994) Dynamics of cadherin/catenin complex formation: novel protein interactions and pathways of complex assembly. J Cell Biol 125, 1327-1340.
38. Young CS, Kitamura M, Hardy S, Kitajewski J (1998) Wnt-1 induces growth, cytosolic β-catenin, and Tcf/Lef transcriptional activation in Rat-1 fibroblast. Mol Cell Biol 18, 2474-2485.
39. Olmeda D, Castel S, Vilaró S, Cano A (2003) β-Catenin regulation during the cell cycle: implications in G2/M and apoptosis. Mol Biol Cell 14, 2844-2860.
40. Lee WA (2005) Prognostic significance of abnormal β-catenin expression in breast carcinoma. Korean J Pathol 39, 114-119.
41. Kallakury BV, Sheehan CE, Winn-Deen E, Oliver J, Fisher HA, Kaufman RP Jr, Ross JS (2001) Decreased expression of catenins (α and β), p120 CTN, and E-cadherin cell adhesion proteins and E-cadherin gene promoter methylation in prostatic adenocarcinomas. Cancer 92, 2786-2795.
42. Odajima T, Sasaki Y, Tanaka N, Kato-Mori Y, Asanuma H, Ikeda T, Satoh M, Hiratsuka H, Tokino T, Sawada N (2005) Abnormal β-catenin expression in oral cancer with no gene mutation: correlation with expression of cyclin D1and epidermal growth factor receptor, Ki-67 labelling index, and clinicopathological features. Hum Pathol 36, 234-241.